

Mesenchymal Stem Cell Classification during Differentiation Based on Shape Information

Introduction

Regenerative therapy using the patient's stem cells is a promising approach for the regeneration of the locomotory system, e.g. bone, cartilage and the intervertebral disc¹. "Stemness" (ability to differentiate into various tissue types) of *in vitro* cultures is difficult to predict due to the heterogeneous nature of cell populations. Here, we investigate whether commitment of stem cells can be tracked using descriptors of shape extracted from 2D time-lapse phase-contrast microscopy data.

Materials and Methods

Human bone marrow was harvested from patients undergoing hip or spine surgery. The procedure was approved by the local Ethics Office. Human mesenchymal stem cells (hMSCs) were amplified from "buffy coat" after density gradient centrifugation by selection for plastic adherence. Passage 3 cells were kept in inductive media for osteogenic, adipogenic, myogenic, 3D chondrogenic (not live tracked) and control and kept in culture for 11 days. The time-lapse imaging was conducted with IncuCyte Plus® (Essen Instruments). Cell phenotype during differentiation was monitored by real time RT-PCR analysis of key genes.

A segmentation pipeline was implemented in order to identify groups of cells. To divide groups into individual cells we used Poisson equation thresholding as described in² where the authors used this algorithm to identify human fingers. Here, we took advantage of the analogy between human fingers and cell "fingers". The feature space in which we performed statistical classification analysis was 8-dimensional: Area, major and minor axis length, perimeter, eccentricity, extent, diameter, number of fingers and day. The classification was performed using decision trees.

Results

In Fig. 1, a pruned version of the classification tree is depicted. The tree shows that 82 % of the

cells that have 10 or more fingers are myogenic progenitors.

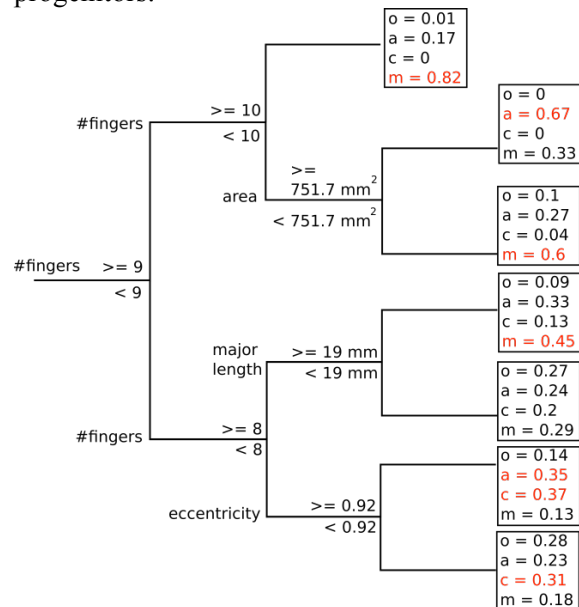


Fig. 1. Classification tree: Class probability for (o)steogenic, (a)dipogenic, (m)yogenic and undifferentiated (c)ontrol at the leaves.

Discussion and Conclusions

The classification results suggest that the four examined cell types can be divided into two groups: Myogenic and non-myogenic cell types, with a classification error of 21.6 %. The presented classification is purely based on shape, other descriptors might be necessary to classify other differentiation groups.

References

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2. Gorelick L, Galun M, Sharon E, Basri R and Brandt A. (2006) Shape Representation and Classification Using the Poisson Equation. *IEEE TPAMI*, **28**(12):1991-2005.

Disclosures

The authors have nothing to disclose.